

\* \* \* \* \* STN Columbus \* \* \* \* \*

FILE 'HOME' ENTERED AT 16:47:28 ON 09 JAN 2004

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COST IN U.S. DOLLARS

SINCE FILE

TOTAL

ENTRY

SESSION

FULL ESTIMATED COST

0.21

0.21

FILES 'MEDLINE, SCISEARCH, LIFESCI, BIOTECHDS, BIOSIS, EMBASE, HCAPLUS, NTIS,  
ESBIOBASE, BIOTECHNO, WPIDS' ENTERED AT 16:47:54 ON 09 JAN 2004  
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11 FILES IN THE FILE LIST

=> s (commercial or scale or batch) (10a) (sialyl? or glycosylat?)

FILE 'MEDLINE'

38986 COMMERCIAL

120028 SCALE

9420 BATCH

6543 SIALYL?

39942 GLYCOSYLAT?

L1 56 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'SCISEARCH'

87545 COMMERCIAL

278198 SCALE

33844 BATCH

6705 SIALYL?

30960 GLYCOSYLAT?

L2 88 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'LIFESCI'

21315 COMMERCIAL

32165 SCALE

10276 BATCH

1615 SIALYL?

9593 GLYCOSYLAT?

L3 27 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'BIOTECHDS'

5741 COMMERCIAL

14335 SCALE

12107 BATCH

403 SIALYL?

3526 GLYCOSYLAT?

L4 66 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'BIOSIS'

85230 COMMERCIAL

139389 SCALE

22994 BATCH

7315 SIALYL?

34885 GLYCOSYLAT?

L5 73 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'EMBASE'

36480 COMMERCIAL

131776 SCALE

15222 BATCH

6182 SIALYL?

31840 GLYCOSYLAT?

L6 75 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'HCAPLUS'  
28321 COMMERCIAL  
266795 COM  
280438 COMMERCIAL  
(COMMERCIAL OR COM)  
314173 SCALE  
78225 BATCH  
8086 SIALYL?  
37716 GLYCOSYLAT?  
L7 144 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'NTIS'  
51928 COMMERCIAL  
80947 SCALE  
6305 BATCH  
18 SIALYL?  
118 GLYCOSYLAT?  
L8 1 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'ESBIOBASE'  
20071 COMMERCIAL  
49588 SCALE  
10033 BATCH  
2703 SIALYL?  
11895 GLYCOSYLAT?  
L9 41 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'BIOTECHNO'  
14938 COMMERCIAL  
23003 SCALE  
11409 BATCH  
3202 SIALYL?  
16990 GLYCOSYLAT?  
L10 46 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

FILE 'WPIDS'  
39606 COMMERCIAL  
117188 SCALE  
26215 BATCH  
410 SIALYL?  
2412 GLYCOSYLAT?  
L11 12 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

TOTAL FOR ALL FILES  
L12 629 (COMMERCIAL OR SCALE OR BATCH) (10A) (SIALYL? OR GLYCOSYLAT?)

=> s l12 not 1998-2004/py

FILE 'MEDLINE'  
2979269 1998-2004/PY  
L13 29 L1 NOT 1998-2004/PY

FILE 'SCISEARCH'  
5861045 1998-2004/PY  
L14 45 L2 NOT 1998-2004/PY

FILE 'LIFESCI'  
617203 1998-2004/PY  
L15 15 L3 NOT 1998-2004/PY

FILE 'BIOTECHDS'  
103353 1998-2004/PY  
L16 47 L4 NOT 1998-2004/PY

FILE 'BIOSIS'

3254024 1998-2004/PY  
 L17 42 L5 NOT 1998-2004/PY  
  
 FILE 'EMBASE'  
 2634784 1998-2004/PY  
 L18 44 L6 NOT 1998-2004/PY  
  
 FILE 'HCAPLUS'  
 5491477 1998-2004/PY  
 L19 74 L7 NOT 1998-2004/PY  
  
 FILE 'NTIS'  
 117194 1998-2004/PY  
 L20 1 L8 NOT 1998-2004/PY  
  
 FILE 'ESBIOBASE'  
 1701387 1998-2004/PY  
 L21 12 L9 NOT 1998-2004/PY  
  
 FILE 'BIOTECHNO'  
 724097 1998-2004/PY  
 L22 24 L10 NOT 1998-2004/PY  
  
 FILE 'WPIDS'  
 4772772 1998-2004/PY  
 L23 3 L11 NOT 1998-2004/PY  
  
 TOTAL FOR ALL FILES  
 L24 336 L12 NOT 1998-2004/PY

=> dup rem l24  
 PROCESSING COMPLETED FOR L24  
 L25 156 DUP REM L24 (180 DUPLICATES REMOVED)

=> d tot

L25 ANSWER 1 OF 156 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN  
 TI Control of interferon-gamma **glycosylation** by the addition of  
 defined lipid supplements to **batch** cultures of recombinant  
 Chinese hamster ovary cells.  
 SO Funatsu, K. [Editor]; Shirai, Y. [Editor]; Matsushita, T. [Editor]. (1997)  
 pp. 339-345. Animal Cell Technology.  
 Publisher: Kluwer Academic Publishers, PO Box 989, 3300 AZ Dordrecht,  
 Netherlands; Kluwer Academic Publishers, 101 Phillip Drive, Norwell,  
 Massachusetts 02061, USA. Series: Animal Cell Technology.  
 Meeting Info.: Eighth Annual Meeting of the Japanese Association for  
 Animal Cell Technology. Iizuka, Japan. November 6-10, 1995.  
 ISBN: 0-7923-4486-3.  
 AU Green, N. H. [Reprint author]; Hooker, A. D. [Reprint author]; James, D.  
 C. [Reprint author]; Baines, A. J. [Reprint author]; Strange, P. G.;  
 Jenkins, N.; Bull, A. T. [Reprint author]  
 AN 1997:466359 BIOSIS  
  
 L25 ANSWER 2 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI Chemoenzymatic synthesis of GM3, Lewis-X and sialyl-Lewis-X  
 oligosaccharides in 13C-enriched form;  
 ganglioside-GM3 oligosaccharide etc. production by sialylation with  
 Trypanosoma cruzi recombinant trans-sialidase, and fucosylation with  
 milk fucosyltransferase  
 SO Tetrahedron Lett.; (1997) 38, 33, 5861-64  
 CODEN: TELEAY ISSN: 0040-4039  
 AU Probert M A; Milton M J; Harris R; Schenkman S; Brown J M; Homans S W;  
 \*Field R A  
 AN 1997-10511 BIOTECHDS

L25 ANSWER 3 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 1  
 TI Gamma-interferon production and quality in stoichiometric fed-batch  
 cultures of Chinese hamster ovary (CHO) cells under serum-free conditions  
 SO BIOTECHNOLOGY AND BIOENGINEERING, (5 DEC 1997) Vol. 56, No. 5, pp.  
 577-582.  
 Publisher: JOHN WILEY & SONS INC, 605 THIRD AVE, NEW YORK, NY 10158-0012.  
 ISSN: 0006-3592.  
 AU Xie L Z; Nyberg G; Gu X J; Li H Y; Mollborn F; Wang D I C (Reprint)  
 AN 97:814754 SCISEARCH

L25 ANSWER 4 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI Production of recombinant proteins in transgenic plants: practical  
 considerations;  
 a review  
 SO Biotechnol.Bioeng.; (1997) 56, 5, 473-84  
 CODEN: BIBIAU ISSN: 0006-3592  
 AU Kusnadi A R; \*Nikolov Z L; Howard J A  
 AN 1998-00604 BIOTECHDS

L25 ANSWER 5 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 2  
 TI Production of recombinant human antithrombin III on 20-L bioreactor scale:  
 Correlation of supernatant neuraminidase activity, desialylation, and  
 decrease of biological activity of recombinant glycoprotein  
 SO BIOTECHNOLOGY AND BIOENGINEERING, (20 NOV 1997) Vol. 56, No. 4, pp.  
 441-448.  
 Publisher: JOHN WILEY & SONS INC, 605 THIRD AVE, NEW YORK, NY 10158-0012.  
 ISSN: 0006-3592.  
 AU Munzert E; Heidemann R; Bunttemeyer H; Lehmann J; Muthing J (Reprint)  
 AN 97:789543 SCISEARCH

L25 ANSWER 6 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Prospective randomized study comparing the efficacy of bioequivalent doses  
 of glycosylated and nonglycosylated rG-CSF for mobilizing peripheral blood  
 progenitor cells  
 SO British Journal of Haematology (1997), 96(2), 418-420  
 CODEN: BJHEAL; ISSN: 0007-1048  
 AU De Arriba, F.; Lozano, M. L.; Ortuno, F.; Heras, I.; Moraleda, J. M.;  
 Vicente, V.  
 AN 1997:177333 HCAPLUS  
 DN 126:220508

L25 ANSWER 7 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 3  
 TI Site- and branch-specific sialylation of recombinant human  
 interferon-gamma in Chinese hamster ovary cell culture  
 SO BIOTECHNOLOGY AND BIOENGINEERING, (20 JUL 1997) Vol. 55, No. 2, pp.  
 390-398.  
 Publisher: JOHN WILEY & SONS INC, 605 THIRD AVE, NEW YORK, NY 10158-0012.  
 ISSN: 0006-3592.  
 AU Gu X J; Harmon B J; Wang D I C (Reprint)  
 AN 97:515441 SCISEARCH

L25 ANSWER 8 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Three types of recombinant human granulocyte colony-stimulating factor  
 have equivalent biological activities in monkeys  
 SO Cytokine (1997), 9(5), 360-369  
 CODEN: CYTIE9; ISSN: 1043-4666  
 AU Tanaka, Hideji; Tanaka, Yoshihiro; Shinagawa, Kyoko; Yamagishi, Yuji;  
 Ohtaki, Kenji; Asano, Katsuhiko  
 AN 1997:400352 HCAPLUS  
 DN 127:117775

L25 ANSWER 9 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 4  
 TI Influence of Primatone RL supplementation on sialylation of recombinant

- human interferon-gamma produced by Chinese hamster ovary cell culture using serum-free media
- SO BIOTECHNOLOGY AND BIOENGINEERING, (20 NOV 1997) Vol. 56, No. 4, pp. 353-360.  
 Publisher: JOHN WILEY & SONS INC, 605 THIRD AVE, NEW YORK, NY 10158-0012.  
 ISSN: 0006-3592.
- AU Gu X J; Xie L Z; Harmon B J; Wang D I C (Reprint)  
 AN 97:789535 SCISEARCH
- L25 ANSWER 10 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Control of interferon-gamma **glycosylation** by the addition of defined lipid supplements to **batch** cultures of recombinant Chinese hamster ovary cells
- SO Animal Cell Technology: Basic & Applied Aspects, Proceedings of the Annual Meeting of the Japanese Association for Animal Cell Technology, 8th, Fukuoka, November 6-10, 1995 (1997), Meeting Date 1995, 339-345.  
 Editor(s): Funatsu, Kazumori; Shirai, Yoshihito; Matsushita, Taku.  
 Publisher: Kluwer, Dordrecht, Neth.  
 CODEN: 64WUA2
- AU Green, N. H.; Hooker, A. D.; James, D. C.; Baines, A. J.; Strange, P. G.; Jenkins, N.; Bull, A. T.  
 AN 1997:563298 HCAPLUS  
 DN 127:219334
- L25 ANSWER 11 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI High resolution glycoform analysis of recombinant human interferon-gamma during batch cultures of Chinese hamster ovary cells
- SO Animal Cell Technology: Basic & Applied Aspects, Proceedings of the Annual Meeting of the Japanese Association for Animal Cell Technology, 8th, Fukuoka, November 6-10, 1995 (1997), Meeting Date 1995, 315-321.  
 Editor(s): Funatsu, Kazumori; Shirai, Yoshihito; Matsushita, Taku.  
 Publisher: Kluwer, Dordrecht, Neth.  
 CODEN: 64WUA2
- AU Hooker, A. D.; Goldman, M. H.; Green, N. H.; James, D. C.; Bull, A. T.; Strange, P. G.; Baines, A. J.; Jenkins, N.  
 AN 1997:563295 HCAPLUS  
 DN 127:219333
- L25 ANSWER 12 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 5  
 TI Purification and characterization of an alpha-L-rhamnosidase from *Aspergillus niger*
- SO FEMS MICROBIOLOGY LETTERS, (15 DEC 1997) Vol. 157, No. 2, pp. 279-283.  
 Publisher: ELSEVIER SCIENCE BV, PO BOX 211, 1000 AE AMSTERDAM, NETHERLANDS.  
 ISSN: 0378-1097.
- AU Manzanares P; deGraaff L H; Visser J (Reprint)  
 AN 1998:201218 SCISEARCH
- L25 ANSWER 13 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI Purification and characterization of an alpha-L-rhamnosidase from *Aspergillus niger*; enzyme purification
- SO FEMS Microbiol.Lett.; (1997) 157, 2, 279-83  
 CODEN: FMLED7 ISSN: 0378-1097
- AU Manzanares P; de Graaff L H; \*Visser J  
 AN 1998-01164 BIOTECHDS
- L25 ANSWER 14 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Epitope determination for antibodies raised against recombinant human interferon-gamma
- SO Animal Cell Technology: From Vaccines to Genetic Medicine, [Proceedings of the Meeting of the ESACT], 14th, Vilamoura, Port., May 1996 (1997), Meeting Date 1996, 277-282. Editor(s): Carrondo, Manuel J. T.; Griffiths, Bryan; Moreira, Jose L. P. Publisher: Kluwer, Dordrecht, Neth.

CODEN: 64ELAL

AU Hooker, Andrew D.; Green, Nicola H.; James, David C.; Strange, Philip G.;  
Baines, Anthony J.; Bull, Alan T.; Jenkins, Nigel

AN 1997:222311 HCAPLUS

DN 126:249970

  

L25 ANSWER 15 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN

TI Chromatographic determination of extinction coefficients of  
non-glycosylated proteins using refractive index (RI) and UV absorbance  
(UV) detectors: applications for studying protein interactions by size  
exclusion chromatography with light-scattering, UV, and RI detectors

SO Techniques in Protein Chemistry VIII, [Symposium of the Protein Society],  
10th, San Jose, Aug. 3-7, 1996 (1997), Meeting Date 1996, 113-119.  
Editor(s): Marshak, Daniel R. Publisher: Academic, San Diego, Calif.  
CODEN: 65GDAE

AU Wen, Jie; Arakawa, Tsutomu; Wypych, Jette; Langley, Keith E.; Schwartz,  
Meredith G.; Philo, John S.

AN 1997:721366 HCAPLUS

DN 128:32085

  

L25 ANSWER 16 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN

TI Sialylation of interferon- $\gamma$  in chinese hamster ovary cell culture

SO Book of Abstracts, 213th ACS National Meeting, San Francisco, April 13-17  
(1997), BIOC-106 Publisher: American Chemical Society, Washington, D. C.  
CODEN: 64AOAA

AU Gu, Xuejun; Wang, Daniel I. C.

AN 1997:230172 HCAPLUS

  

L25 ANSWER 17 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN

TI Synthesis and structural characterization of an antibacterial  
glycoprotein.

SO Book of Abstracts, 214th ACS National Meeting, Las Vegas, NV, September  
7-11 (1997), CARB-066 Publisher: American Chemical Society, Washington, D.  
C.  
CODEN: 64RNAO

AU Winans, K. A.; King, D. S.; Bertozzi, C. R.

AN 1997:486022 HCAPLUS

  

L25 ANSWER 18 OF 156 MEDLINE on STN DUPLICATE 6

TI Chemoenzymatic synthesis of a trimeric ganglioside GM3 analogue.

SO CARBOHYDRATE RESEARCH, (1997 Jun 11) 301 (1-2) 1-4.  
Journal code: 0043535. ISSN: 0008-6215.

AU Earle M A; Manku S; Hultin P G; Li H; Palcic M M

AN 97372515 MEDLINE

  

L25 ANSWER 19 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

TI Sialylation of interferon-gamma in Chinese hamster ovary cell culture;  
recombinant protein preparation in CHO cell culture (conference  
abstract)

SO Abstr.Pap.Am.Chem.Soc.; (1997) 213 Meet., Pt.1, BIOT106  
CODEN: ACSRAL ISSN: 0065-7727  
American Chemical Society, 213th ACS National Meeting, San Francisco, CA,  
13-17 April, 1997.

AU Gu X; Wang D I C

AN 1997-11606 BIOTECHDS

  

L25 ANSWER 20 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

TI Baculo virus vectors comprising a signal peptide and promoter;  
for e.g. recombinant HIV virus-1 gp120 overexpression and  
glycosylation in a Spodoptera frugiperda Sf9 insect cell culture

AU Murphy C I; Young E

AN 1996-08666 BIOTECHDS

PI US 5516657 14 May 1996

L25 ANSWER 21 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI Enzymatic galactosylation of sugars with in situ regeneration of  
 nucleotide sugar;  
 oligosaccharide production with coenzyme regeneration, using  
 sucrose-synthase, beta-1,4-galactosyltransferase and  
 UDP-glucose-4-epimerase, with activator addition  
 AU Hoersch B; Seiffert-Storiko A; Marquardt R; Zervosen A; Elling L; Kula M  
 R  
 AN 1997-01028 BIOTECHDS  
 PI WO 9635801 14 Nov 1996

L25 ANSWER 22 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 8  
 TI A CONVENIENT AND EFFICIENT SYNTHESIS OF SLEX ANALOGS  
 SO JOURNAL OF ORGANIC CHEMISTRY, (03 MAY 1996) Vol. 61, No. 9, pp. 2938-2945.  
 ISSN: 0022-3263.  
 AU HAYASHI M (Reprint); TANAKA M; ITOH M; MIYAUCHI H  
 AN 96:363122 SCISEARCH

L25 ANSWER 23 OF 156 MEDLINE on STN DUPLICATE 9  
 TI Oligosaccharide mapping reveals hormone-specific glycosylation patterns on  
 equine gonadotropin alpha-subunit Asn56.  
 SO ENDOCRINOLOGY, (1996 Jun) 137 (6) 2543-57.  
 Journal code: 0375040. ISSN: 0013-7227.  
 AU Gotschall R R; Bousfield G R  
 AN 96217295 MEDLINE

L25 ANSWER 24 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 10  
 TI A new assay for sialyltransferases using fluorescein-labeled acceptors  
 SO Liebigs Annalen (1996), (11), 1773-1784  
 CODEN: LANAEM; ISSN: 0947-3440  
 AU Limberg, Gerrit; Slim; George C.; Compston, Catherine A.; Stangier, Peter;  
 Palcic, Monica M.; Furneaux, Richard H.  
 AN 1997:149544 HCAPLUS  
 Correction of: 1996:678653  
 DN 126:154367  
 Correction of: 126:16187

L25 ANSWER 25 OF 156 MEDLINE on STN DUPLICATE 11  
 TI Exploring the substrate specificities of alpha-2,6- and  
 alpha-2,3-sialyltransferases using synthetic acceptor analogues.  
 SO EUROPEAN JOURNAL OF BIOCHEMISTRY, (1996 Dec 15) 242 (3) 674-81.  
 Journal code: 0107600. ISSN: 0014-2956.  
 AU Van Dorst J A; Tikkanen J M; Krezdorn C H; Streiff M B; Berger E G; Van  
 Kuik J A; Kamerling J P; Vliegthart J F  
 AN 97175036 MEDLINE

L25 ANSWER 26 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Large-scale preparation of sialyloligosaccharide from  
 egg yolk  
 SO Baiošaiensu to Indasutori (1996), 54(3), 200-1  
 CODEN: BIDSE6; ISSN: 0914-8981  
 AU Koketsu, Mamoru; Juneja, Lekh Raj; Kim, Mujo  
 AN 1996:335752 HCAPLUS  
 DN 125:5709

L25 ANSWER 27 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Synthesis and biological evaluation of clitocine analogs as adenosine  
 kinase inhibitors.  
 SO Book of Abstracts, 212th ACS National Meeting, Orlando, FL, August 25-29  
 (1996), MEDI-156 Publisher: American Chemical Society, Washington, D. C.  
 CODEN: 63BFAF  
 AU Lee, Chih-Hung; Jiang, Meiqun; Daanen, Jerry; Kohlaas, Kathy L.;  
 Alexander, Karen M.; Yu, Haixia; Kowaluk, Elizabeth A.; Bhagwat, Shripad  
 S.

AN 1996:414744 HCAPLUS

L25 ANSWER 28 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN  
 TI **GLYCOSYLATION** ANALYSIS OF A MURINE MONOCLONAL-ANTIBODY DURING  
**SCALE-UP** FROM ROLLER BOTTLE TO 200L FERMENTER  
 SO ABSTRACTS OF PAPERS OF THE AMERICAN CHEMICAL SOCIETY, (24 MAR 1996) Vol.  
 211, Part 1, pp. 133-BIOT.  
 ISSN: 0065-7727.  
 AU BHAT R (Reprint); JOHNSON L; MEIDER P; KELSEY W  
 AN 96:249930 SCISEARCH

L25 ANSWER 29 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI **Glycosylation** analysis of a murine monoclonal antibody during  
**scale-up** from roller bottle to 200L fermentor  
 SO Book of Abstracts, 211th ACS National Meeting, New Orleans, LA, March  
 24-28 (1996), BIOT-133 Publisher: American Chemical Society, Washington,  
 D. C.  
 CODEN: 62PIAJ  
 AU Bhat, Ramadas; Johnson, L.; Meider, P.; Kelsey, W.  
 AN 1996:217290 HCAPLUS

L25 ANSWER 30 OF 156 MEDLINE on STN DUPLICATE 12  
 TI Enlarged **scale** chemical synthesis and range of activity of  
 drosocin, an O-**glycosylated** antibacterial peptide of Drosophila.  
 SO EUROPEAN JOURNAL OF BIOCHEMISTRY, (1996 May 15) 238 (1) 64-9.  
 Journal code: 0107600. ISSN: 0014-2956.  
 AU Bulet P; Urge L; Ohresser S; Hetru C; Otvos L Jr  
 AN 96248422 MEDLINE

L25 ANSWER 31 OF 156 MEDLINE on STN DUPLICATE 13  
 TI Separation of human serum transferrin isoforms by high-performance  
 pellicular anion-exchange chromatography.  
 SO PROTEIN EXPRESSION AND PURIFICATION, (1996 Feb) 7 (1) 39-44.  
 Journal code: 9101496. ISSN: 1046-5928.  
 AU Rohrer J S; Avdalovic N  
 AN 96209930 MEDLINE

L25 ANSWER 32 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
 14  
 TI HIGH-LEVEL SECRETION OF HUMAN ALPHA(1)-ANTITRYPSIN FROM  
 SACCHAROMYCES-CEREVISIAE USING INULINASE SIGNAL SEQUENCE  
 SO JOURNAL OF BIOTECHNOLOGY, (18 JUL 1996) Vol. 48, No. 1-2, pp. 15-24.  
 ISSN: 0168-1656.  
 AU KANG H A; NAM S W; KWON K S; CHUNG B H; YU M H (Reprint)  
 AN 96:689358 SCISEARCH

L25 ANSWER 33 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI **Glycosylation** analysis of a murine monoclonal antibody during  
**scale-up** from roller bottle to 200-l fermentor;  
 mouse hybridoma cell culture in a perfusion culture vessel (conference  
 abstract)  
 SO Abstr.Pap.Am.Chem.Soc.; (1996) 211 Meet., Pt.1, BIOT133  
 CODEN: ACSRAL ISSN: 0065-7727  
 211th ACS National Meeting, New Orleans, CA, 24-28 March, 1996.  
 AU Bhat R; Johnson L; Meider P; Kelsey W  
 AN 1996-08743 BIOTECHDS

L25 ANSWER 34 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI Alpha-2,8-sialyltransferase cDNA;  
 gene cloning and expression; DNA probe for tumor-associated antigen  
 detection and cancer diagnosis  
 AN 1996-03350 BIOTECHDS  
 PI JP 07327678 19 Dec 1995



L25 ANSWER 35 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
 17  
 TI STERIC CONTROL OF N-ACETYLGALACTOSAMINE IN GLYCOSIDIC BAND FORMATION  
 SO TETRAHEDRON LETTERS, (18 SEP 1995) Vol. 36, No. 38, pp. 6839-6842.  
 ISSN: 0040-4039.  
 AU YULE J E; WONG T C; GANDHI S S; QIU D X; RIOPEL M A; KOGANTY R R (Reprint)  
 AN 95:656984 SCISEARCH

L25 ANSWER 36 OF 156 MEDLINE on STN DUPLICATE 18  
 TI Large-scale expression of recombinant **sialyltransferases**  
 and comparison of their kinetic properties with native enzymes.  
 SO GLYCOCONJUGATE JOURNAL, (1995 Dec) 12 (6) 755-61.  
 Journal code: 8603310. ISSN: 0282-0080.  
 AU Williams M A; Kitagawa H; Datta A K; Paulson J C; Jamieson J C  
 AN 96318012 MEDLINE

L25 ANSWER 37 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
 19  
 TI AN IMPROVED PROCEDURE FOR THE SYNTHESIS OF 1,3-DIDEAZAADENOSINE  
 SO SYNTHETIC COMMUNICATIONS, (1995) Vol. 25, No. 5, pp. 711-718.  
 ISSN: 0039-7911.  
 AU DEVLIN T A; JEBARATNAM D J (Reprint)  
 AN 95:138669 SCISEARCH

L25 ANSWER 38 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
 20  
 TI THE INFLUENCE OF DIETS AND GUT MICROFLORA ON LECTIN-BINDING PATTERNS OF  
 INTESTINAL MUCINS IN RATS  
 SO LABORATORY INVESTIGATION, (OCT 1995) Vol. 73, No. 4, pp. 558-564.  
 ISSN: 0023-6837.  
 AU SHARMA R (Reprint); SCHUMACHER U  
 AN 95:751628 SCISEARCH

L25 ANSWER 39 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 TI Effect of lipids on recombinant interferon- $\gamma$  glycosylation  
 SO Animal Cell Technology: Developments towards the 21st Century,  
 [Proceedings of the Meeting], Veldhoven, Neth., Sept. 12-16, 1994 (1995),  
 Meeting Date 1994, 391-396. Editor(s): Beuvery, E. Coen; Griffiths, J.  
 Brian; Zeijlemaker, Wim P. Publisher: Kluwer, Dordrecht, Neth.  
 CODEN: 62VAAP  
 AU Jenkins, Nigel; Castro, Paula M. L.; Menon, Sunitha; Ison, Andrew P.;  
 Bull, Alan T.  
 AN 1996:307959 HCAPLUS  
 DN 124:340584

L25 ANSWER 40 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 TI Glycosylation heterogeneity of recombinant plasminogen expressed in CHO  
 cells;  
 determined using capillary zone electrophoresis and matrix-assisted  
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L25 ANSWER 87 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
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L25 ANSWER 94 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN

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L25 ANSWER 95 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

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L25 ANSWER 104 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
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L25 ANSWER 111 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

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L25 ANSWER 124 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
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L25 ANSWER 128 OF 156 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN  
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L25 ANSWER 130 OF 156 MEDLINE on STN DUPLICATE 54  
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L25 ANSWER 131 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN  
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L25 ANSWER 132 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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 CODEN: PIXXD2  
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	W: JP, SU				
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L25 ANSWER 134 OF 156 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS  
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L25 ANSWER 135 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 139 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 140 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 141 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
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L25 ANSWER 142 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
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L25 ANSWER 144 OF 156 MEDLINE on STN DUPLICATE 58



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L25 ANSWER 145 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 146 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 147 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 149 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 150 OF 156 MEDLINE on STN DUPLICATE 59  
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L25 ANSWER 152 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
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L25 ANSWER 153 OF 156 MEDLINE on STN DUPLICATE 60  
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L25 ANSWER 154 OF 156 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN  
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L25 ANSWER 155 OF 156 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN  
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L25 ANSWER 156 OF 156 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS  
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ANSWER SET L25 HAS BEEN SAVED AS 'BATCH/A'

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L25 ANSWER 5 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 2  
 AB Chinese hamster ovary (CHO) cells producing the recombinant  
 glycoprotein human antithrombin III (rhAT III) were batch cultivated in a  
 20-L bioreactor for 13 days. Neuraminidase activity in cell-free  
 supernatant was monitored during cultivation and free sialic acid was  
 determined by HPLC. Neu5Ac alpha(2-->3)Gal-specific Maackia amurensis and  
 Gal beta(1-->4)GlcNAc-specific Datura stramonium agglutinin were used for  
 determination of **sialylated** and desialylated rhAT III,

respectively. A **commercial** test kit was used for evaluation of functional rhAT III activity. Supernatant neuraminidase as well as lactate dehydrogenase activity increased significantly during batch growth. The enhanced number of dead cells correlated with increased neuraminidase activity, which seemed to be principally due to cell lysis, resulting in release of cytosolic neuraminidase. Loss of terminally alpha(2-->3) linked sialic acids of the oligosaccharide portions of rhAT III, analyzed in lectin-based Western blot and lectin-adsorbent assays, correlated with a decrease of activity of rhAT III produced throughout long-term batch cultivation. Thus, structural oligosaccharide integrity as well as the functional activity of recombinant glycoprotein depend on the viability and mortality of the bioreactor culture, and batches with a high number of viable cells are required to guarantee production of glycoproteins with maximum biological activity. (C) 1997 John Wiley & Sons, Inc.

L25 ANSWER 9 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 4

AB Although serum-free media have been widely used in mammalian cell culture for therapeutic protein production, the effects of serum-substitutes on product quality have not been extensively examined. This study observed an adverse effect of Primatone RL, an animal tissue hydrolysate commonly used as a serum-substitute to promote cell growth, on sialylation of interferon-gamma (IFN-gamma) derived from Chinese hamster ovary (CHO) cell culture in both **batch** and **fed-batch** modes. In **batch** cultures, decreased **sialylation** was observed at each of the **glycosylation** sites (i.e., Asn(25) and Asn(97)) of IFN-gamma with the use of elevated concentrations of the peptone. Although poorest sialylation was obtained with the use of a growth-inhibiting concentration of Primatone RL, diminished sialylation was observed at the optimal peptone concentration for cell growth and product yield. Since incubation of the product in Primatone RL-supplemented acellular medium did not result in decreased sialylation, the negative effect of Primatone RL could not be attributed to extracellular desialylation of IFN-gamma by components of the peptone. In the fed-batch mode, a culture utilizing a serum-free feeding medium supplemented with Primatone RL demonstrated poorer sialylation than a similar culture not fed the peptone. The results of both the batch and fed-batch experiments indicate that the adverse effect of the peptone was not due solely to ammonia accumulation. (C) 1997 John Wiley & Sons, Inc.

L25 ANSWER 19 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB Sialylation of interferon-gamma (IFN-gamma) produced by CHO cell culture was monitored by reverse-phase HPLC separations of the site-specific pools of tryptic glycopeptides representing the product's 2 potential N-linked glycosylation sites (i.e. Asn25 and Asn97). The IFN-gamma displayed both site- and branch-specific differences in sialylation as the Asn25 site and the Man(alpha-1,3) branch of the predominant complex biantennary glycan structures at each site were preferentially sialylated. When the **sialylation** profile of IFN-gamma was analyzed throughout a suspension **batch** culture, **sialylation** at each site and branch was found to be incomplete but relatively constant until a steady decrease in sialylation was observed concurrent with loss of cell viability. The introduction of competitive sialidase-inhibitor into the culture supernatant prevented the loss of sialylation following but not prior to cell death, thus indicating that the sialic acid content of the final product was determined by both incomplete intracellular sialylation and extracellular desialylation. The influences of culture medium on sialylation were also studied. (0 ref)

L25 ANSWER 21 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB A new method for enzymatic galactosylation of a monosaccharide or oligosaccharide, with in situ nucleotide sugar coenzyme regeneration, involves reaction with sucrose-synthase (SS, EC-2.4.1.13), beta-1,4-galactosyltransferase (GT) and UDP-glucose-4-epimerase (UDPGP,

EC-5.1.3.2), and a keto sugar or derivative is added as an activator of UDPGE. The activator is preferably dUDP-6-deoxy-D-xylohexulose, TDP-6-deoxy-D-xylo-hexulose, 6-deoxyglucosone, galactosone, allosone or glucosone, at 0.01-20 (preferably 0.1) mM, and the process is carried out as a repetitive batch operation in an ultrafiltration cell. The products are useful as precursors of sialylated or fucosylated sugars, e.g. for production of sialyl-Lewis-X or derivatives involved in cell-cell recognition. The activator reactivates UDPGE in situ, eliminating the need for repeated addition of this expensive enzyme and allowing repeated use without immobilization, and has no adverse effects on activity of the other enzymes. (36pp)

L25 ANSWER 26 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN

AB A review, with 10 refs., on the structure and a large-scale preparation of **sialyloligosaccharides** from egg yolk.

L25 ANSWER 28 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN

L25 ANSWER 30 OF 156 MEDLINE on STN DUPLICATE 12

AB Insects respond to a bacterial challenge by rapidly synthesizing a diverse range of antibacterial and antifungal peptides. One of them, drosocin, a 19-residue proline-rich antibacterial peptide, was isolated from *Drosophila*. This peptide carries a disaccharide moiety attached to a threonine residue in mid-chain position. The present report describes the enlarged-scale chemical synthesis of drosocin, **glycosylated** with Gal (beta 1 --> 3)GalNAc(alpha 1 --> O). We have studied the range of activity of the synthetic glycopeptide, of two truncated glycosylated isoforms, and of the unglycosylated L and D enantiomers. Both isolated and chemically synthesized drosocins carrying the disaccharide display the same antibacterial activity. Using circular dichroic spectroscopy we demonstrated that the O-linked disaccharidic motif did not affect the backbone conformation of drosocin. The antibacterial activity of the synthetic glycopeptide was directed against gram-negative strains with the exception of the gram-positive bacteria *Micrococcus luteus*. Deletion of the first five N-terminal residues completely abolished the activity of drosocin. As a first approach to the study of the mode of action of drosocin, we have synthesized a non-glycosylated D enantiomer and, using this molecule, we have shown that drosocin may act on the gram-negative bacteria through a stereospecific target.

L25 ANSWER 41 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 21

AB A Chinese hamster ovary cell line expressing recombinant human interferon-gamma (IFN-gamma) was grown in a 15-l stirred tank fermenter. N-linked carbohydrate populations associated with both Asn(25) and Asn(97) were isolated by reverse-phase HPLC separation of trypsin-digested IFN-gamma and their structure determined by matrix-assisted laser desorption/ionisation mass spectrometry (MALDI-MD) in combination with exoglycosidase array sequencing. The predominant oligosaccharide at both glycosylation sites throughout the culture was a complex biantennary structure, Gal(2)GlcNAc(2)Man(3)GlcNAc(2), which was fucosylated when attached to Asn(25) but not to Asn(97). A gradual decrease in this biantennary structure was observed, with a concomitant increase in the proportion of truncated and high-mannose glycans. These experiments demonstrate the relative stability of **glycosylation** during **batch** culture and the definitive site-specific **glycosylation** data that can be obtained using MALDI-MS as a monitoring technique.

L25 ANSWER 42 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB The control of N-glycosylation in a CHO-320 cell line that produces recombinant interferon-gamma (IFN-G) was used as a model system to study the effects on the glycosylation process of protein structure, host cell

type and cell culture conditions. CHO-320 cells were adapted to grow suspended in serum-free medium based on RPMI-1640 supplemented with cattle serum albumin (CSA), insulin, transferrin and trace element supplements. The glycoform proportions were held constant at steady-state using glucose-limited chemostat systems; at a constant dilution rate of 0.015/hr, cell growth and IFN-G were transiently improved by pulses of 3.8 mM or 5.0 mM glucose. The lipoprotein supplement ExCyte minimized **glycosylation** deterioration in **batch** culture, and partially substituted the CSA content of the medium with a fatty acid-free preparation had a similar effect. Recombinant IFN-G was routinely purified from cell culture supernatant using an anti-IFN-G immunoaffinity matrix, yielding more than 98% pure IFN-G. Oligosaccharide structures of CHO cell-derived IFN-G, the influence of host cell type on IFN-G glycosylation, and the consequences of drug efficacy were also discussed. (28 ref)

- L25 ANSWER 43 OF 156 MEDLINE on STN DUPLICATE 22  
 AB The culture environment exerts a major effect on the glycosylation pattern of recombinant human interferon-gamma (IFN-gamma) produced by Chinese-hamster ovary (CHO) cells. The recombinant IFN-gamma is heterogeneous and consists of a mixture of fully (2N), partially (1N) and non-**glycosylated** (0N) glycoforms, and throughout **batch** cultures there is a decline in the proportion of fully **glycosylated** IFN-gamma. Glucose and glutamine, nutrients that are depleted early in such cultures, were *prima facie* candidates for causing such a shift in glycoform profile. Batch feeding of these nutrients did not prevent the decline in 2N glycoform, but the glycosylation pattern of IFN-gamma was affected by the initial glutamine concentration in the culture. Under different serum-free environments the extent of IFN-gamma glycosylation was affected by (1) the concentration of BSA, (2) the quality of BSA, (3) the lipid composition of the culture medium and (4) the presence of surfactants. Moreover, the inclusion of serum in cultures caused changes in the molecular masses of the major glycoforms, that was indicative of cleavage of the core polypeptide. The results reported emphasize the necessity of considering the effects of culture media on product quality as well as on product quantity during process optimization.
- L25 ANSWER 44 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 AB Engineering issues in applied mammalian cell culture are reviewed with respect to: (1) secreted products from mammalian cells (monoclonal antibody production as a model for secreted protein production, increasing the productivity of monoclonal antibody production, product quality issues, and cell culture vessels for the production of secreted mammalian cell products); (2) mammalian cells as products (factors controlling cell growth and differentiation, and culture vessel design for stem cell expansion). Engineering principles which have been applied to fermentor designs for microbial systems may be used to create mammalian cell culture vessels for quite dissimilar applications. For protein expression challenges remain in increasing specific productivity of cell lines, enabling protein-free culture on a large-**scale** and the control of quality aspects e.g. **glycosylation** heterogeneity and viral clearance. In stem cell culture, challenges remain in improving recovery during cell separation, media design and culture vessel design to ensure tight control of important factors (dissolved oxygen tension). (21 ref)
- L25 ANSWER 45 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN  
 AB Gal $\beta$ 1,4GlcNAc  $\alpha$ 2,6-sialyltransferase (EC 2.4.99.1) catalyzes the incorporation of sialic acids at the terminal positions of glycoconjugates through a NeuAc  $\alpha$ 2,6-Gal linkage. The cDNA sequences for mouse, rat, human and chicken, along with the genomic DNA sequence, and tissue specific alternative splicing in rat have been reported. To gain a further insight into the structure and function

relationship, we attempted the large **scale** production of a recombinant **sialyltransferase** in *Escherichia coli* in an insol. form. The product was solubilized with urea, and renatured to give the active enzyme. The renatured enzyme was similar to the enzyme obtained from rat liver, except for its dependence on ionic strength.

L25 ANSWER 47 OF 156 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE  
23

AB Fed-batch culture currently represents the most attractive choice for large scale production of monoclonal antibodies (MAbs), due to its operational simplicity, reliability, and flexibility for implementation in multipurpose facilities. Development of highly productive cell lines, maximization of cell culture longevity, and maintenance of high specific antibody secretion rates through genetic engineering techniques, nutrient supplementation, waste product minimization, and control of environmental conditions are important for the design of high-yield fed-batch processes. Initially simple supplementation protocols have evolved into sophisticated serum-free multivitamin feeds that result in MAb titers on the order of 1-2 g/L. Limited research has been published to date on the effects of various culture parameters on potentially important quality issues, such as MAb **glycosylation** and stability. Although most fed-batch protocols to date have relied on relatively simple control schemes, increasingly sophisticated algorithms must be applied in order to take full advantage of the potentially additive effects of manipulating nutrient and environmental parameters to maximize fed-batch process productivity.

L25 ANSWER 48 OF 156 NTIS COPYRIGHT 2004 NTIS on STN

AB Specificity determinants of human acetylcholinesterase (HuAChE) towards ligands (substrate and some reversible and irreversible inhibitors) were identified by combination of site-directed mutagenesis, molecular modeling and kinetic studies with enzymes mutated in active center residues Trp86, Glu202, Trp286, Phe295, Phe297, Tyr337, Phe338 and Glu450. Thus, the anionic and hydrophobic subsites as well as the acyl pocket were identified. Enzymes with resistance to OP aging were engineered. The role of N-glycosylation in the function, biosynthesis and stability of HuAChE was examined by site-directed mutagenesis (Asn to Gln substitution) of the three potential N **glycosylation** sites, Asn265, Asn350 and Asn464. Large **scale** preparation of recombinant HuAChE was performed utilizing the microcarrier technology. Over 500 milligrams of enzyme was prepared for x-ray crystallography.

L25 ANSWER 55 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB Recombinant protein glycosylation is reviewed with respect to: (1) reasons why **glycosylation** is significant (**commercial** importance, methods used to study effects of **glycosylation**, and the effects on protein solubility, protein stability, biological activity, pharmacokinetics and immunogenicity); (2) oligosaccharide structures (N-glycosylation and O-glycosylation); (3) glycan analysis (electrophoresis, chromatography, NMR and MS); and (4) influences on glycosylation (protein structure, host cell type, culture environment and method of cell culture). Improvements in analytical procedures offer detailed glycan analysis during or soon after host cell culture. Detailed knowledge of glycoprotein biosynthesis may aid control of glycan heterogeneity by using culture media formulations/supplements. Host cells may be engineered for biased production of certain glycoforms. Advances in carbohydrate chemistry and recombinant glycosyltransferases may lead to construction of complex oligosaccharides, which may be grafted onto recombinant proteins made in prokaryotes. Until this time, human recombinant glycoproteins are best produced in animal cell culture. (165 ref)

L25 ANSWER 70 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB Glycosylation is cell type-specific, and thus recombinant proteins

produced in heterologous systems are almost invariably glycosylated differently from the native form. Differences can include differences in both the number of attached glycan chains and precise glycan sequences at an individual glycosylation site. Glycosylation can influence activity, pharmacokinetics, and immunogenicity, and different glycosylation patterns may be associated with differences in the therapeutic profile. It is thus useful to analyze the glycosylation pattern of a recombinant protein at as early a stage as possible, and to compare to the native form, and to screen for determinants which interact with the immune system and lectins. The glycosylation pattern is very sensitive to culture method and variations in the extracellular environment, and it is thus important during **scale-up** to ensure that the **glycosylation** pattern is maintained. A production process is only valid if it reproducibly allows isolation of protein with a constant **glycosylation** pattern. It is useful to assess **glycosylation** on a **batch-to-batch** basis. (0 ref)

L25 ANSWER 72 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 AB Separation of a glycosylated 28-residue synthetic peptide from byproducts of the glycosylation reaction was performed by displacement chromatography in a reverse-phase system with benzyldimethylhexadecylammonium chloride as the displacer and a water/acetonitrile/phosphoric acid system. During method development using a column of internal diameter 0.46 cm problems attributed to either adsorption azeotropy or aggregation were overcome by optimizing acetonitrile concentration and operating at 55 deg. The method was scaled-up to 22 g per run on an axial compression column of internal diameter 15 cm. Compared with conventional elution chromatography conducted on a similar scale, the displacement process realized a nearly 8-fold increase in throughput with a significant reduction in solvent consumption. Details regarding process development and scale-up were presented. (0 ref)

L25 ANSWER 76 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 AB Applications of aldolases and transferases in production of sialic acid derivatives were discussed. Sialyl-aldolase was used for production of 3-deoxy-D-manno-2-octulosonic acid. N-glycolylneuraminic acid (NeuGc) was produced on a large **scale** from N-glycolylglycosamine using **sialyl**-aldolase, and CMP-NeuGc (a tumor-associated antigen precursor) was produced using acylneuraminate-cytidylyltransferase (EC-2.7.7.43). 2 Pig liver sialyltransferase enzymes were purified by affinity chromatography, immobilized and used in sialyloligosaccharide production. Alpha-2,6-galactosyl- beta-1,4-N-acetylglucosamine-sialyltransferase was used for sialylation of 3 different synthetic oligosaccharides. Alpha-2,3-galactosyl- beta-1,3-N-acetylglactosamine-sialyltransferase reacted with Gal-beta-1,3-GlcNAc, giving the 1st preparative synthesis of NeuAc-alpha-2,3-Gal-beta-1,3-GlcNAc (an epitope of the human pancreas adenocarcinoma tumor-associated antigen CA-50). Enzymatic synthesis seems to be the method of choice for modification of oligosaccharide structures on glycoproteins. (5 ref)

L25 ANSWER 81 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN  
 AB The recent development of enzyme-catalyzed reactions for the production of sugars, peptides and related substances was discussed. Topics considered included: the preparation of uncommon and aza sugars by aldolase-catalyzed aldol condensation followed by Pd-mediated reductive amination; methods for enzyme-catalyzed **glycosylation** using glycosyltransferase, glycosidase, transglycosidase and phosphorylase enzymes; large-**scale** production of oligosaccharides catalyzed by glycosyltransferases with in situ regeneration of sugar nucleotides; the coupling of glycosidase- and glycosyltransferase-catalyzed reactions for oligosaccharide production with minimal requirements for sugar nucleotide regeneration; cloning and expression of the catalytic domain

of glycosyltransferase for oligosaccharide production in *Escherichia coli*; the glycosyltransferase-catalyzed production of uncommon oligosaccharides such as sialyl Lewis x and sialyl Le(x) glycol; production of large peptides and their conjugates; the use of enzyme engineering to make enzymes more stable in dimethylformamide; and engineering subtilisin (EC-3.4.21.14) to catalyze ligation reactions. (58 ref)

L25 ANSWER 83 OF 156 HCAPLUS COPYRIGHT 2004 ACS on STN

AB Recombinant human interleukin-5 (hIL-5) has been expressed at high levels and produced in large quantities in baculovirus infected Sf9 insect cells. The glycosylated protein was purified using immuno-affinity chromatog. and gel filtration. Purified hIL-5 has been crystallized using standard vapor diffusion techniques with PEG as a co-precipitant. The crystals belong to the C2 space group and diffract to 2 Å.

L25 ANSWER 91 OF 156 MEDLINE on STN DUPLICATE 42

AB alpha-Neup5Ac-(2----3)-beta-D-Galp-(1----3)-D-GlcpNAc (2) and, alpha-Neup5Ac-(2----3)-beta-D-Galp-(1----3)-beta-D-GlcpNAcOMBn++ were prepared on a large scale by the action of beta-D-Galp-(1----3)-D-GalpNAc (2----3)-alpha-sialyltransferase (partially purified from porcine liver) on beta-D-Galp-(1----3)-D-GlcpNAc and beta-D-Galp-(1----3)-beta-D-GlcpNAcOMBn, respectively. The trisaccharide 2 is the epitope of the tumor-associated carbohydrate antigen CA 50, highly expressed in human pancreatic adenocarcinoma.

L25 ANSWER 96 OF 156 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN

L25 ANSWER 104 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB Human recombinant interleukin-3 (IL-3) was produced by gene cloning and expression in bacteria (*Escherichia coli* HB101, DH1, MC1061, *Bacillus subtilis* 1A40 and *Bacillus licheniformis* T9), yeast (*Saccharomyces cerevisiae* D273-10B and *Kluyveromyces lactis* CBS 2360) and mammalian cells (COS, C127 (ATCC CRL 1616) and CHO-12). A low-cost production and purification scheme was designed using *B. licheniformis* because the protein secreted by *B. licheniformis* was not glycosylated and had a mol. weight of about 15,000. Vector plasmid pGB/IL-322 and plasmid pGB/IL-326 were constructed containing the alpha-amylase (EC-3.2.1.1) signal peptide fused to the sequences encoding mature IL-3 and placed downstream of a strong alpha-amylase and *HpaII* promoter, respectively. IL-3 (3 g) was purified from cell-free filtrate (48 l) by hydrophobic interaction chromatography on Fractogel TSK butyl 650C, 60% (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub> precipitation, anion-exchange chromatography on Q-Sepharose Fast Flow and concentration by ultrafiltration (twice), gel filtration on Sephacryl S100HR and ultrafiltration. The purified and formulated product entered clinical trials in November, 1989. (28 ref)

L25 ANSWER 119 OF 156 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

AB A human cDNA containing the complete coding region for the lysosomal glycoprotein glucocerebrosidase (EC-3.2.1.45) was introduced into the genome of *Autographa californica* nuclear-polyhedrosis virus (AcNPV) downstream from the polyhedrin promoter. The recombinant virus (pAc373/GC) was cotransfected with wild-type AcNPV DNA into *Spodoptera frugiperda* SF9 cells using a modified calcium phosphate precipitation technique. Recombinant baculo virus containing human glucocerebrosidase cDNA was obtained, and this was plaque-purified and used to infect SF9 cells. The recombinant enzyme was characterized and found to be active in SF9 cells. High levels of glucocerebrosidase were produced; 40% of which was in the culture medium. The N-terminal amino acid sequence of the recombinant product was identical to that of mature, human placental glucocerebrosidase. The enzyme in the culture supernatant and in the SF9 cells was **glycosylated**. The insect cell culture system could be used for large-scale recombinant glucocerebrosidase production, which is of clinical interest. (48 ref)



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